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SUBJECT—

TITLE

RELATION OF UNSEASONAL TEMPERATURES TO BARK-BEETLE MORTALITY

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INTRODUCTION

The study of temperatures lethal to overwintering broods of the mountain pine beetle instituted at the Coeur d'Alene laboratory in the fall of 1936 has been continued during each subsequent winter. Results obtained from experiments conducted during the winters of 1936-37 and 1937-38 have been presented in unpublished laboratory reports.*

Although during these two seasons a great deal of work was conducted on the different phases of the problem and in developing an operative technique, the conclusions drawn from these experiments can be summarized in the following statement: When mountain pine beetle broods are subjected to normal fall and winter conditions, sufficient cold-hardiness is developed to withstand the minimum winter temperatures of the northern Rocky Mountain Region. However, the occurrence of abnormal or unseasonal temperatures at a time when sufficient cold-hardiness has not been developed will result in comparable degrees of

* Low temperatures critical to bark-beetle larvae, by James C. Evenden. The effect of tempering as a means of increasing cold-hardiness upon the lipid and moisture content of mountain pine beetle larvae, by W. D. Bedard. July 9, 1937

* The relation of unseasonal temperatures to bark-beetle mortality, by James C. Evenden. The relation of lipid and moisture content to cold-hardiness of mountain pine beetle larvae, by W. D. Bedard. June 24, 1938

brood mortality. Such unseasonal conditions exist in extreme low temperatures in the fall prior to the development of resistance, or in the spring after cold-hardiness has been broken. It is also possible for unusually warm temperatures during winter months to reduce the cold-hardiness of overwintering bark beetle broods to a degree that mortality is associated with subsequent normal seasonal conditions.

EXPERIMENTS OF 1938-39 SEASON

Experiments of the previous season were not entirely satisfactory, as the minimum temperature (-30° F.) that could be obtained with the equipment at the Coeur d'Alene laboratory was not sufficient to produce mortality in overwintering broods of the mountain pine beetle during periods of maximum cold-hardiness. During the winter of 1938-1939 the work was planned so as to determine if possible the maximum larval resistance and to check the seasonal development of cold-hardiness.

During the period of maximum cold-hardiness when the minimum temperature of the laboratory equipment was insufficient to cause mortality, decreased temperatures were obtained by placing "dry ice" in the cabinet. As such temperatures were below the functional limits of the thermostatic control of the equipment, an electric light was placed in the temperature chamber and used as a heater to maintain the desired condition. By checking with potentiometer reading at frequent intervals and switching the light on and off a desired temperature could be maintained within a range of ± 1° F.

DETERMINATION OF MORTALITY FOLLOWING EXPOSURES TO SET TEMPERATURES

The technique of operation necessary in satisfying the immediate requirements of this study has not been above just criticism. Although

in transfering the hibernating larvae from the logs to the petri dishes for subsequent exposure care is exercised so as to avoid marked changes from normal conditions, there is undoubtedly some shock associated with this operation. It is unreasonable to assume that the placing of the naked larvae within the paraffin cells of the petri dishes reestablishes natural environmental conditions. Although the above objections are fully appreciated, perhaps the greatest weakness in the operative technique rested in determining the actual mortality following an exposure to a critical temperature. The practice followed in determining mortality was to allow the solidly frozen larvae to warm gradually and under proper humidity conditions. The material is then examined at 24-, 48-, and 72-hour intervals after exposure. At each of these examinations the active and normally colored larvae were removed from the petri dishes as having withstood the exposure. Larvae which were dormant at the 24- and 48-hour examination were often distinctly alive and active at the 72-hour examination. Although there is no question but that these larvae were alive, it is reasonable to assume that such extreme exposures would result in some internal injury that would affect subsequent normal development.

Attempts were made to determine the extent of this delayed or post-exposure mortality, without a great deal of success. Prepupal larvae that were frozen solid at various temperatures ultimately recovered and pupated; however, it is not known if they developed into normal adults. Different methods of attempting to rear the exposed larvae were employed, but there were so many variations in the results obtained that no conclusions could be drawn. As a result of the complex

association of factors connected with these tests the data obtained are considered as showing only the seasonal development of cold-hardiness, and the extent to which the larvae can at least temporarily recover from an exposure to freezing temperatures. Although the relationship between the immediate recovery and actual development to normal adults is a problem yet to be solved, one may safely assume that fully resistant mountain pine beetle larvae from northern Idaho can successfully withstand temperatures of at least -35° F.

1938-1939 EXPERIMENTS

DEVELOPMENT OF COLD-HARDINESS

To determine the seasonal development of the cold-hardiness of mountain pine beetle larvae, a series of tests were started in August 1938 and repeated at stated intervals until May 1939. The same technique as used during previous experiments was employed in these tests. Petri dishes containing fifty larvae were exposed to set temperatures for periods of 2 hours and 15 minutes. Although these different tests were planned to include the range of lethal temperatures, in some few instances an error was made in determining the point at which mortality started. Data obtained from these tests are shown in chart I.

On August 27, mountain pine beetle larvae were extremely susceptible to low temperatures. The occurrence of a period of unseasonal or abnormal low temperature at that time would have resulted in severe brood mortality. With the occurrence of cold fall temperatures larval resistance developed until in February a maximum cold-hardiness was reached.

On October 10 the data obtained are sufficiently out of line to call for some explanation as to the reduced cold-hardiness. The larvae used for this test were from the same source as those used on September 24 and October 24. On October 10 the weather was moderate with a heavy warm rain falling at the time the larvae were collected. Although active when collected, it is possible that the larval resistance was still further reduced by being transported from the field to the laboratory in wet frass, which was a condition that had not been anticipated and for which protection had not been provided. The conditions surrounding this test were not desirable; however, it does serve as an illustration of the degree to which the cold-hardiness of mountain pine beetle larvae can fluctuate under changed climatic conditions.

The variation of cold-hardiness as developed by individual larvae is portrayed by these data. This is not entirely due to a difference in larval development, as a marked variation occurs with larvae of the same instar, although prepupal larvae do show a greater resistance. A number of factors such as larval development, moisture content, body chemistry, protection afforded by different bark thickness, and possibly others, must be responsible for this variation in the cold-hardiness of mountain pine beetle larvae.

An improper representation of the different degrees of cold-hardiness within the 50 larvae used for each of these tests explains some of the irregularities of the data as submitted. These irregularities are illustrated by the October 24 data, where a 34 percent mortality followed an exposure to +5° F. and only a 32 percent mortality was associated with a +0.5° F. exposure.

At no time do all larvae, even in the same tree, reach the same degree of cold-hardiness, although their resistance apparently increases proportionately with the duration of midwinter temperatures. The data obtained on December 12 show a lag in the resistance of individual larvae, and would indicate that under all conditions some larvae will be susceptible to temperatures considerably higher than that required to produce a 100 percent mortality.

Data obtained on January 12 show the extreme temperatures employed during this series of tests. The larvae which withstood these exposures were placed within small cells in the phloem of a fresh white pine slab. On January 22, a number of the larvae from the -61° F. exposure had done some feeding; however, they soon became inactive and died. The larvae from the -78° F. exposure lived until January 22, but did no feeding. This test was carried one step further than shown by these data with larvae being exposed at -116° F. and although 68 percent of them recovered and became active they all died prior to January 22. Although the ultimate mortality of larvae removed from their natural hibernating conditions and exposed to such extreme temperatures would seem to be assured, these data can not be interpreted as depicting the existence of cold-hardiness under normal conditions.

Chart II gives a range of temperatures critical to mountain pine beetle larvae from white pine during the winter of 1938-1939. The curve depicting the temperature at which no mortality occurred is a smoothed curve plotted on the highest temperature points.

Monthly minimum air temperatures from Coeur d'Alene, Idaho, are compared to the trend of these curves to record the relation of cold-hardiness development to existing weather conditions. It is realized

that these data do not represent conditions in the area where the infested logs were stored, but they do depict the general trend of temperatures for the area. The data obtained from these tests did not permit the extremes of these curves to be joined.

A sharp increase in larval resistance is shown in the 100 percent mortality curve for May 1939. This increase resulted from the occurrence of prepupal larvae, as at the time the material for the May 15 test was collected a fairly large percent of the brood had pupated.

PROLONGED EXPOSURES TO DECREASING TEMPERATURES

It has been previously shown* that prolonged exposures to a set temperature will not increase larval mortality. For example, mountain pine beetle larvae exposed for 10 days at -25° F. showed no significant difference in mortality from those exposed to the same temperature for 2 hours.

The following test was planned to determine if a prolonged exposure to a constantly lowering temperature would result in an increased mortality. Nine petri dishes of 50 mountain pine beetle larvae to each dish were placed in the cabinet at a temperature of +26° F. at 1:20 p.m. January 4, and lowered to -50° F. on January 6, 1939. As the temperature of the cabinet was lowered a dish was removed at stated intervals and subjected to the same treatment and examination as previously stated. The results of this test are shown in the following table:

* The relation of unseasonal temperatures to bark-beetle mortality, by James C. Evenden. The relation of lipid and moisture content to cold-hardiness of mountain pine beetle larvae, by W. D. Bedard. June 24, 1938.

TABLE I

COMPARISON OF MOUNTAIN PINE BEETLE LARVAL MORTALITY
RESULTING FROM PROLONGED EXPOSURE TO DECREASING TEMPERATURES
AND FROM A TWO HOUR EXPOSURE AT A SET TEMPERATURE
EXPERIMENT STARTED AT +26° F.

Date	Time petri dish was removed	Hours of exposure	Temperature when dish was removed	Percent of larval mortality	Mortality resulting from 2-hour exposure on January 12, 1939
January 4,1939	11:15 p.m.	9 hr. 55 min.	- 8° F.	0	—
January 5, 1939	11:22 p.m.	22 hr. 2 min.	-15° F.	0	—
	3:50 p.m.	26 hr. 30 min.	-19.5° F.	0	2
	10:40 p.m.	33 hr. 20 min.	-26° F.	2	4
January 6,1939	9:27 a.m.	44 hr. 07 min.	-33° F.	16	14
	12:05 p.m.	46 hr. 45 min.	-35° F.	16	14
	2:50 p.m.	49 hr. 30 min.	-40° F.	16	28
	5:23 p.m.	52 hr. 03 min.	-45° F.	24	26
	8:20 p.m.	56 hr. 00 min.	-50° F.	24	26

From the preceding data it will be seen that the prolonged exposure of mountain pine beetle larvae to a decreasing temperature did not increase the mortality associated with each exposure. It becomes apparent that after the body of the larvae has been chilled the severity of the temperature and not the length of exposure determines its lethal properties.

COLD-HARDINESS OF MOUNTAIN PINE BEETLE ADULTS

As a comparison of the cold-hardiness of mountain pine beetle new adults and larvae, the following test was conducted on September 24, 1938. It was intended to continue these comparative tests at different times during the winter, but insufficient adult material within the stored logs did not permit of this plan. The technique employed in this test was the same as used for larvae. New adults and larvae were exposed to set temperatures for a period of 2 hours and 15 minutes. The results of this test are shown in table II.

TABLE II

COMPARISON OF THE COLD-HARDINESS OF MOUNTAIN PINE LARVAE AND ADULTS WHEN EXPOSED TO SAME TEMPERATURES

Date	Temperature during exposure +20	Adult mortality 0	Larval mortality 0
September 24, 1939	+15	0	2%
	+10	32%	94%
	+ 6	32%	92%
	+1	44%	90%
	- 3	46%	100%
	- 8	84%	100%
	-13	100%	100%
	-17	100%	100%

From the data in table II it would appear that on the date that this test was conducted the new adults had developed a higher degree of cold-hardiness. However, it is realized that this one test is not significant to permit the drawing of conclusions.

COLD-HARDINESS OF MOUNTAIN PINE BEETLE UNDER NATURAL WINTER HIBERNATION CONDITIONS

To test the resistance of mountain pine beetle larvae under natural winter hibernating conditions, infested white pine logs were subjected to severe low temperatures. Previous tests of this character had shown that very little brood mortality followed exposure to temperatures of -25° F. during periods of maximum larval cold-hardiness. These exposures were of sufficient duration (10 to 30 hours) to lower the temperature beneath the bark to that of the cabinet. These experiments were controlled from a section of each log that was removed and held as a check. Of the three tests made, the adult emergence from the exposed logs and checks was 61 - 62, 26 - 12, and 101 - 100

respectively. These results make it apparent that such an exposure is not sufficient to cause any abnormal mortality.

In tests conducted during the winter of 1938-1939, dry ice was used, which provided temperatures of -25° F., -30° F., and -42° F. beneath the infested bark of the exposed logs. Although the same technique was used in these tests as previously employed, the exposed logs molded quite badly within the emergence cages, and even in the -25° F. exposure only 2 mountain pine beetle, a few secondary beetles and several parasites emerged. Only 2 mountain pine beetle adults emerged from the logs exposed to -30° F., and none from the -42° F. exposure. Although no positive explanation is available, it is evident that a difference in the moisture content of the logs was responsible for this condition. The only change in the technique as previously employed with satisfaction, was the use of dry ice, but it is doubtful if the gas from this substance would have resulted in the condition as stated. However, for all practical purposes these experiments were a failure.

SUMMARY

Data from the experiments conducted during the winter of 1938-1939 substantiate information obtained from previous tests. They demonstrate the building of an increased larval resistance to a degree which at its maximum will withstand seasonal temperatures of the northern Rocky Mountain region.

The development of this increased cold-hardiness is synchronized with the occurrence of fall and winter temperatures, the severity of which governs the degree of resistance attained.

The data obtained also show that during periods of maximum cold-hardiness mountain pine beetle larvae from this region can withstand, at least temporarily, extreme temperatures as low as -60° F. or more. It is appreciated that the exposure of these insects, even under normal hibernating conditions, to such extreme and abnormal temperatures will undoubtedly result in ultimate death although the frozen insects may show a temporary recovery.

These tests also show that the length of exposure does not increase the lethal properties of any set temperature.

A comparison of the cold-hardiness of mountain pine beetle adults and larvae showed the former to be the most resistant. Although one test can not be considered as conclusive, the results were as expected, and would no doubt have been substantiated by additional data.

The information obtained from this series of experiments gives added support to the contention that has come from the work of this laboratory, that overwintering broods of the mountain pine beetle are resistant to normal winter temperatures but susceptible to any unseasonal conditions.

CONCLUSIONS

Recognizing the objections to the technique employed in the execution of these tests, infested logs will be used in all future experiments. This procedure leaves the larvae in natural hibernation conditions, and permits an accurate determination of the true mortality associated with each exposure. Work of this character has been prosecuted during the present winter, but due to the extremely

mild weather that has been experienced a high degree of cold-hardiness was not developed. The results of this winter's tests will not be available until May, when the exposed logs are examined for brood mortality. It is expected that further tests along this line will be necessary during the 1940-41 season.

CHART I

SEASONAL RESISTANCE OF MOUNTAIN PINE BEETLE LARVAE TO LOW TEMPERATURES

WHITE PINE

Larval Mortality Shown in Percent

Date	+20	+10	0	-10	-20	-30	-40	-50	-60	-70
1938 Aug. 27	0	2	20	100						
Sept. 9	0	4	8	88	96	100				
Sept. 24		0	2	94	92	90	100			
Oct. 10		0	60	88	86	96	100			
Oct. 24		0	2	10	34	32	56	76	100	
Nov. 7					0	2	16	52	84	88	92	96				
Nov. 21						0	6	8	10	10	14	18	22	44		
Nov. 28							0	2	4	4	10	8	14	10		
Dec. 12									14	8	12	8	10	22	24	30
1939 Jan. 1									0	2	4	14	14	28	26	26
Jan. 12															8	14
March 6										10	10	34	28			
April 20						0	6	8	32	58	60	70				
April 6			8	36		74	84	98	96	98	98	100				
April 17		0	34	68	76	76	92	100								
May 1	6	22	82	100												
May 15	0	8	26	62	90	96										

CHART II

SEASONAL VARIATIONS IN THE RANGE OF CRITICAL TEMPERATURES FOR MOUNTAIN PINE BEETLE LARVAE IN WHITE PINE 1938 - 1939